Title: Spur Sub-Populations, Fruiting, and Leaf Senescence as a Possible Rationale for the Refinement of Diagnostic Leaf Sampling for Nitrogen in 'Nonpareil' Almond

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Spurs are the fundamental bearing units in mature almond trees and 90% of 'Nonpareil' fruit are borne on spurs (the rest are borne on one year old shoots). Since yield is highly correlated with the number of fruiting spurs, increases in spur death may reduce subsequent yields unless compensated for by the growth of new shoots and spur renewal on those shoots the following year. Spur dynamics have received limited study, but it is believed that spurs only live four to five years. In a previous Almond Board – sponsored study, we reported that 8-10 times more spurs die following a fruiting year than following a year in which they remain vegetative. Moreover, we have documented that surviving fruiting spurs also have fewer flowers (and therefore fruits) in the following spring.

It is clear that within a single almond tree canopy there are distinct spur sub-populations defined not only by differing spur fruiting status, but also by differing spur leaf area or light exposure. Variation among spur sub-populations in floral initiation and mortality rates, as well as leaf changes in those spurs, may provide fresh insights to refine the leaf sampling strategy prior to Nitrogen (N) analyses. Currently, the strategy recommended is to sample leaves randomly from non-fruiting spurs. BY FOCUSING ON FRUITING SPURS, WHICH MAY IMPACT YIELD MORE DIRECTLY THAN THE VEGETATIVE SPURS CURRENTLY SAMPLED, WE MAY DETECT CHANGES IN LEAF N WHICH ARE NOT YET APPARENT IN THE LESS VULNERABLE, NON-FRUITING SPURS.

The biological rationale for our approach is the following: In addition to its role in the photosynthetic apparatus, N in leaves may represent a storage pool which may be utilized by the developing fruit if N availability is limiting—either in the whole tree when soil N availability is inadequate or in more shaded, stressful locations in the tree canopy. Leaf death normally occurs in autumn but, under conditions of N-stress, leaves may die prematurely. In dying leaves, N-containing proteins are broken down and N is translocated to the developing fruit/kernel. Prior to leaf fall, about 50% of the N content of the leaves is translocated elsewhere.

Objectives:

The objective of this work was to establish and test, for increased sensitivity, a diagnostic leaf sampling protocol for leaf N analysis which is founded on tree biological relationships rather than the random sampling protocol employed currently. Not only may leaf analysis become more sensitive, but vulnerable spur sub-populations are likely to be related much more directly to productivity in the subsequent year(s) than the current procedure which focuses on vegetative spurs.

Materials and Methods:

1. **Characterize spur subpopulations as defined by spur fruiting status and light exposure with regards to their vulnerability to N-deprivation.**

Trees growing at Paramount Farms in Lost Hills were subjected to N application rates of either 248 or 124 lbs acre^{-1}. Hundreds of spurs were tagged and monitored from summer 2003 until bloom 2004. Variables considered include the following: specific leaf weight (SLW; leaf dry weight per unit leaf area, an easily measured and reliable indicator of relative leaf light exposure), leaf area per spur, spur survival rates and flower bud number. Additionally, changes in leaf N during kernel growth (a major period of carbohydrate and nitrogen demand in the tree) were examined, indicating the degree, if any, of leaf senescence and N resorption from the leaves of the different spur sub-populations to developing embryos. We plotted the N content and concentration of spur leaves as a function of SLW.

2. **Compare and assess the relative sensitivities to N deprivation of the currently - used and the proposed protocols.**

Using trees subjected to N application rates of either 248 or 124 lbs acre^{-1}, we compared the N concentrations of leaves sampled conventionally with those of leaves sampled from selected vulnerable spur sub-populations to determine whether the vulnerable spurs provide evidence of low N concentration under circumstances in which the conventional protocol is insensitive.

Results and Discussion:

Spur Floral Initiation and Survival.

The death of almond spurs is not random. Fruiting spurs are far less likely to initiate flower buds or survive until spring than are non-fruiting spurs. Also, among fruiting spurs, those with the smallest leaf areas are more likely to die in the winter than those with greater leaf areas per spur. Similarly, flower number per spur increased with increasing leaf area. Assuming leaf area is associated positively with carbohydrate availability (data not presented), these data are consistent with the concept of a relative spur autonomy –at least late in the growing season- with respect to carbohydrate availability. That is, the carbohydrate available for flowering, fruiting and survival of a spur through the process of photosynthesis may be largely restricted to that produced by the leaves OF THAT PARTICULAR SPUR. Adding further support to this notion of spur autonomy, the results of our current experiment demonstrate that, among fruiting spurs, flower bud number and percent spur survival increased with spur light exposure as indicated by increasing SLW (Fig.1 and Table 1). However, because neither flower bud number (data not shown) nor percent survival of spurs (Table 1) were influenced by the nitrogen application treatment, reduced yield due to mild N-stress can not be explained by decreases in spur floral initiation or spur survival rates.

Premature Leaf Drop and Leaf Senescence.

Irrespective of N application rates, about 10% of well-exposed fruiting spurs exhibited premature leaf fall in 2003 (Table 2). Among fruiting spurs in more shaded canopy positions, the percentage of spurs exhibiting premature leaf abscission averaged about 50% higher (about 15% of spurs exhibiting premature leaf fall). These abscission data suggest that fruiting and shading of spurs are associated with increased spur stress and leaf senescence during the period of embryo development.

Relative Sensitivities of Leaf N Sampling Protocols.

Leaf N concentration did not vary with spur light exposure as indicated by SLW (Fig.2). So, at least under the conditions of this experiment, sampling shaded leaves does not provide a more sensitive indicator of tree N-status than does sampling leaves according to the conventional protocol.

Although fruiting spurs had marginally lower leaf N concentrations in July than did non-fruiting spurs (1.89% vs. 2.03% at the higher N application rate and 1.67% vs. 1.80% at the lower N application rate; Table 3A), there was no evidence from leaf N data that the fruiting spur sub-populations were under greater N-stress than the non-fruiting spur sub-populations. Had the fruiting spurs been under greater nitrogen stress than the non-fruiting spurs during embryo development (May to July, 2003) we might have anticipated greater remobilization of spur leaf N to the developing fruits, with possible increases in premature leaf senescence on the fruiting spurs. We did not directly determine the percentage of spur leaf senescence on these spurs; however, it was clear that the decline in leaf N concentration on fruiting spur sub-populations was not any greater than that noted in the non-fruiting spur sub-populations. Thus, the percentage decline in leaf N concentration during the period of embryo development was about 30% in both the fruiting and non-fruiting spur sub-populations on trees receiving 248 lbs N per acre (data not presented). Similarly, fruiting had no impact on the decrease in N concentration in trees receiving 124 lbs N per acre, but the low N application rate was associated with a marginally higher decrease in leaf N concentration of 35% between May and July (data not presented).

We conclude that, in 2003, the fruiting spur sub-populations were not more sensitive to N availability than were the non-fruiting spur sub-populations. It must be noted, however, that differences in the N status of trees receiving 248 lbs N per acre and 124 lbs N per acre were not large (Table 3A). Furthermore, despite N application rates of 248 lbs N per acre, at leaf N concentrations of 2.03% even the "high N" trees were marginally N-deficient (recommended July leaf N concentrations range from 2.2 to 2.5% N). We anticipate that the decrease in leaf N concentrations during embryo development will vary between fruiting and non-fruiting spurs sub-populations as differences in tree N status increase. We expect that in 2004 the "low N" trees will become more N-deficient and the "high N" trees will exhibit leaf N concentrations more typical of well-fertilized trees. In research currently underway we are attempting to impact both soil N availability and tree N demand. Soil N availability is addressed, as previously, as a result of low vs. high fertilizer N application rates. Tree demand for N is being impacted by manipulating tree crop load.

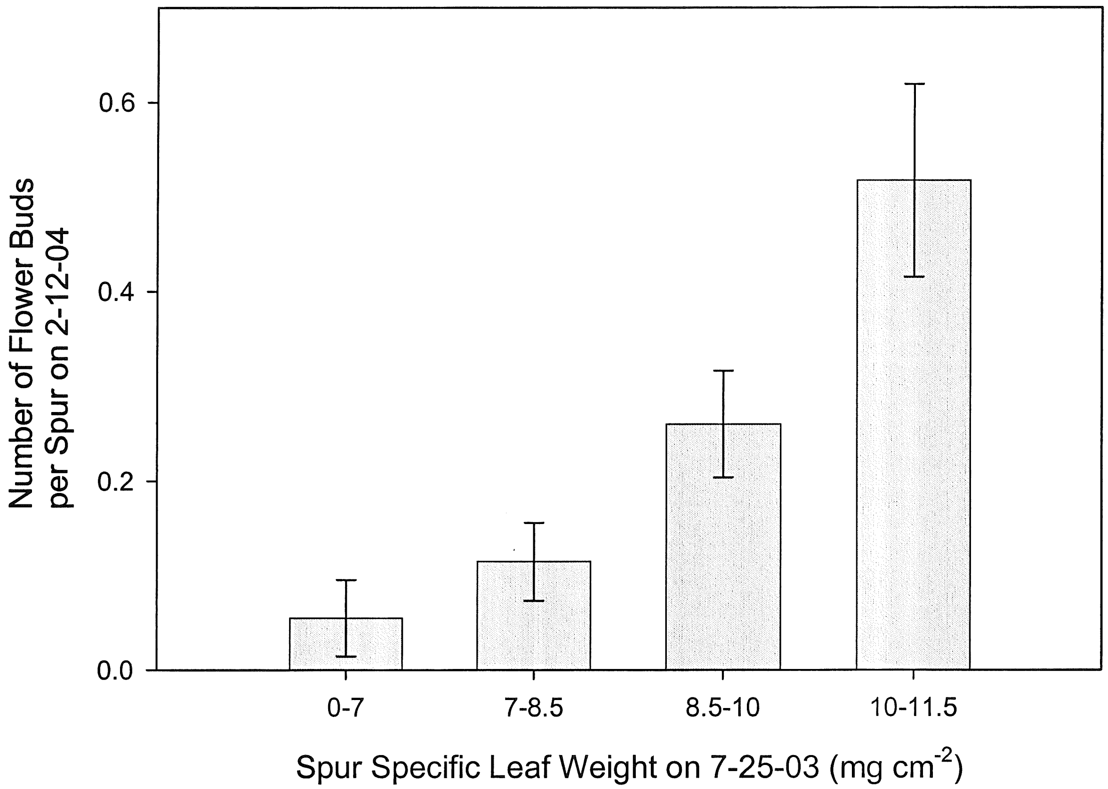

Fig.1 The relationship between light exposure (as indicated by specific leaf weight, i.e. leaf dry weight per unit leaf area) and the number of flower buds number on fruiting spurs in the subsequent spring. Bars indicate ± standard error.

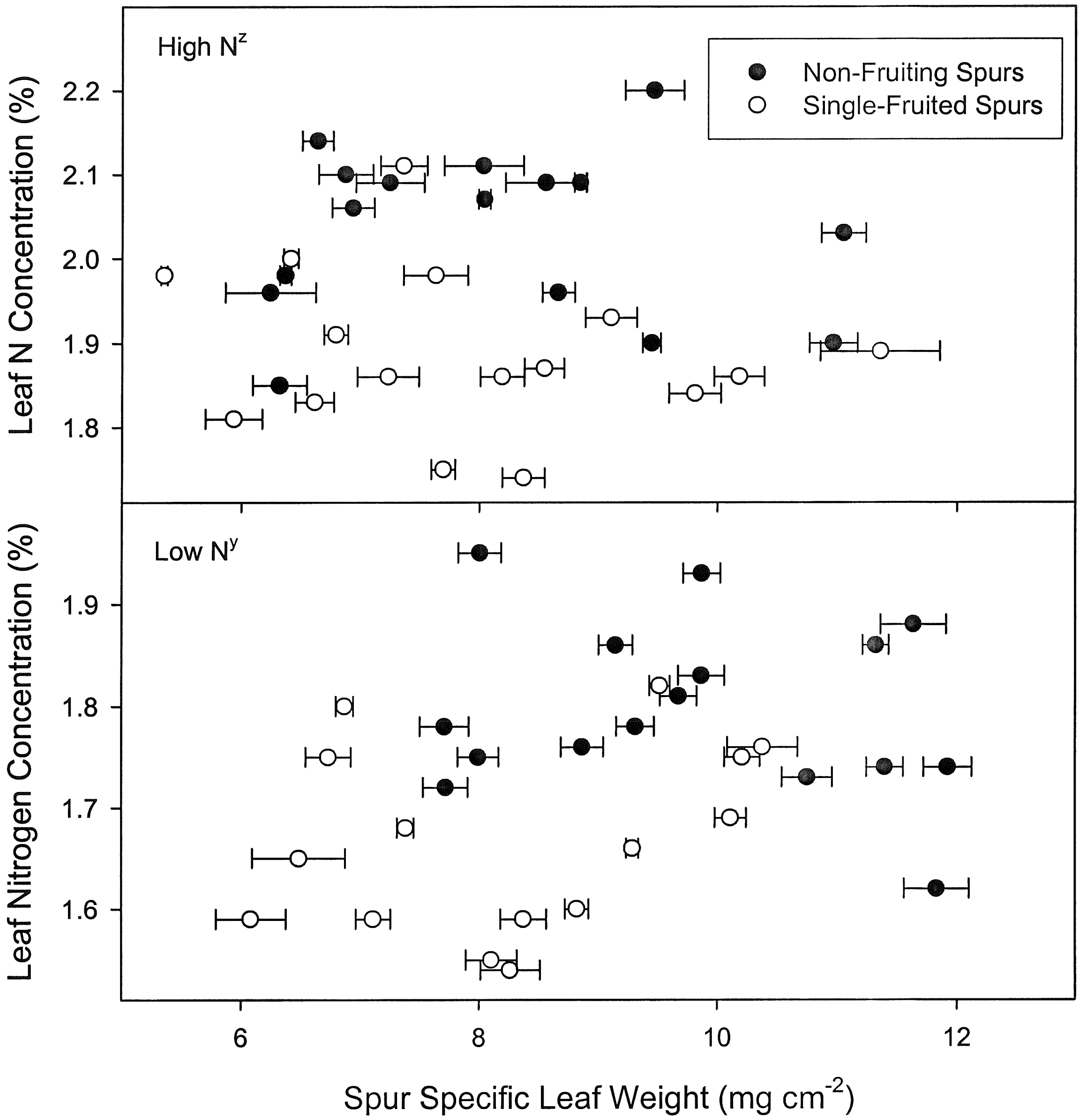

Fig.2 The relationship between spur light exposure (as indicated by specific leaf weight) and spur leaf nitrogen concentration on 7-25-03.

^{z} N applied at 248 lbs acre^{-1} in 2003.

^{y} N applied at 124 lbs acre^{-1} in 2003.

Table 1. The relationship between light exposure (as indicated by specific leaf weight) and subsequent winter mortality rates of fruiting spurs [z].

N Application Rate	Spur Specific Leaf Weight ($mg\ cm^{-2}$) on 7-25-03	Spurs Alive on 2-12-04	Spurs Dead on 2-12-04	Percentage Dead (%)
248 lbs $acre^{-1}$	0-8	41	22	34.9
	8-10	39	4	9.3
	10-12	21	1	4.5
124 lbs $acre^{-1}$	0-8	28	7	20.0
	8-10	53	4	7.0
	10-12	29	1	3.3

[z] Spurs bearing a single fruit in 2003.

Table 2. The relationship between light exposure (as indicated by specific leaf weight) and leaf abscission of fruiting spurs[z] during the period of embryo growth on almond trees subjected to high and low nitrogen application rates.

N Application Rate	Spur Specific Leaf Weight (mg cm^{-2})	Number of Spurs with Leaf Abscission	Number of Spurs With No Leaf Abscission	Percentage of Spurs with Leaf Abscission (%)
248 lbs $acre^{-1}$	5-8.5 (shaded)	10	64	13.5
	8.5-12 (exposed)	5	46	9.8
124 lbs $acre^{-1}$	5-8.5 (shaded)	10	46	17.9
	8.5-12 (exposed)	9	72	11.1

[z] Spurs bearing one fruit in 2003.

Table 3a. The effect of nitrogen application rates on changes in leaf nitrogen concentration of single-fruited and non-fruiting spurs during embryo development (May to July, 2003).

N Application Rate	Spur Fruiting Status (2003)	Leaf N Concentration on 5-7-03 (%)[z]	Leaf N Concentration on 7-25-03 (%)[z]
248 lbs acre^{-1}	Fruiting	2.71 ± 0.07	1.89 ± 0.02
	Non-Fruiting[y]	2.92 ± 0.04	2.03 ± 0.02
124 lbs acre^{-1}	Fruiting	2.59 ± 0.06	1.67 ± 0.02
	Non-Fruiting[y]	2.77 ± 0.04	1.80 ± 0.02

[z] Data are means ± se.
[y] Conventional sample.

Table 3b. The effect of nitrogen application rates on changes in leaf nitrogen per unit leaf area of single-fruited and non-fruiting spurs during embryo development (May to July, 2003).

N Application Rate	Spur Fruiting Status (2003)	Leaf N Per Unit Leaf Area on 5-7-03 (mg cm^{-2})[z]	Leaf N Per Unit Leaf Area on 7-25-03 (mg cm^{-2})[z]
248 lbs acre^{-1}	Fruiting	0.18 ± 0.01	0.15 ± 0.01
	Non-Fruiting[y]	0.21 ± 0.01	0.16 ± 0.01
124 lbs acre^{-1}	Fruiting	0.18 ± 0.01	0.14 ± 0.01
	Non-Fruiting[y]	0.22 ± 0.01	0.18 ± 0.01

[z] Data are means ± se.
[y] Conventional sample.